Variation in Responses of Sunflower Cultivars to the Parasitic Weed Broomrape

H. Eizenberg, D. Plakhine, J. Hershenhorn, and Y. Kleifeld, ARO, Newe Ya'ar Research Center, Ramat Yishay, Israel; and B. Rubin, Faculty of Agriculture, Food, and Environmental Science, The Hebrew University of Jerusalem, Rehovot, Israel

ABSTRACT

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Development of four *Orobanche* species, *O. cumana*, *O. aegyptiaca*, *O. ramosa*, and *O. cernua*, was compared on resistant and susceptible sunflower cultivars. Sunflower plants were infected by *O. cumana*, *O. aegyptiaca*, and *O. ramosa*, but not by *O. cernua*, in field and greenhouse studies. However, cultivating the hosts and parasites in a polyethylene bag system allowed the observation that sunflower induced *O. cernua* seed germination. This difference demonstrates that *O. cernua* is unique from the other three species. *O. cumana*, *O. aegyptiaca*, and *O. ramosa* attached to and developed tubercles on the susceptible sunflower 'Adi.' On the resistant 'Ambar' sunflower, a greater percentage of tubercles were degenerated or dead than on the susceptible cultivar. Thus, resistance of Ambar appears to manifest during tubercle development. Seed production of Adi sunflower was greatly reduced with infection by *O. cumana*.

Additional keywords: Egyptian broomrape, root parasite, sunflower broomrape

The genus *Orobanche* includes chlorophyll-lacking root parasites that parasitize many dicotyledonous species, causing severe damage to vegetable and field crops worldwide (16). *Orobanche cumana*, *O. aegyptiaca*, *O. ramosa*, and *O. cernua* are noxious parasitic weeds that infest most of the agricultural areas in Israel (4,16). Sunflower broomrape (*O. cumana* Wallr.) and Egyptian broomrape (*O. aegyptiaca* Pers.) are the two *Orobanche* spp. that parasitize sunflower (*Helianthus annuus* L.) in Israel (4).

The species *O. cumana* corresponds in Spain to *O. cernua* (1). Katzir et al. (13) confirmed that *O. cumana* and *O. cernua* in Israel are two different species based on random amplified polymorphic DNA (RAPD) markers. In addition, in Israel, the host range of *O. cernua* and *O. cumana* differs: *O. cumana* parasitizes mainly sunflower, whereas *O. cernua* parasitizes mainly species from the *Solanaceae* family (4).

Several crops from various botanical families have been found to be resistant or tolerant to *Orobanche* spp. (1,4). However, an economically significant level of resistance has been developed only in members of the *Fabaceae* family (10) and in sunflower (1,3,6). Field tolerance of parsley to *O. crenata* also has been found in Israel (9). In recent years, resistant oil-sunflower

cultivars were introduced for commercial production in Eastern Europe; however, after a few seasons, they became heavily infected by *O. cumana* (2). In Bulgaria, it was reported that, between 1927 and 1992, five different races of *O. cumana* developed to overcome five different resistances (O_1 to O_5) in oil-sunflower cultivars that were imported from Eastern Europe (7). The same phenomenon was observed in Spain and, recently, two new resistances (O_6 and O_7) were introduced (1).

Several possible mechanisms have been suggested for the resistance phenomenon, such as (i) secretion of inhibitors (phytoalexins) by the resistant host (17), (ii) increased production of phytotoxins in the host roots in response to *Orobanche* spp. penetration (5), and (iii) condensation and lignification of the root cell walls and increased activity of specific enzymes involved in the defense response (8). Several wild sunflower species have been found to be resistant to *O. cumana* because of their ability to increase cell wall deposition or vessel occlusion, or to induce cellular disorganization (14).

In Israel, the resistance of sunflower cv. Ambar to *O. cumana* and *O. aegyptiaca* is temperature dependent (3,6). High temperatures greater than 26 and 18°C (day and night) increase the resistance reaction, whereas low temperatures, lower than 20 and 12°C (day and night), break down the resistance. The resistance manifests as a mortality of the *Orobanche* tubercles after penetration and establishment in the host tissue (3,6). In Spain, sunflower cultivars were highly resistant to *O. cumana* only at 27°C, but were sensitive at the lower temperature range of 15 to 23°C (18).

The objectives of the present study were to (i) compare the responses of resistant and sensitive sunflower cultivars to *O. ramosa* and *O. cernua* infection and to the resistant response that was observed in sunflower infected by *O. cumana* and *O. aegyptiaca*, and (ii) define the host range of *Orobanche* spp. in resistant and sensitive sunflower cultivars in Israel.

MATERIALS AND METHODS

Plant material: sunflower. The resistant (R) Ambar and sensitive (S) Adi sunflower cultivars were provided by Dr. Baruch Retig of the ARO, Israel. Both sunflower cultivars were bred from the local cv. D. Y. 3; therefore, the genetic backgrounds of both cultivars are similar (B. Retig, *personal communication*).

***Orobanche* spp.** *O. cumana* seed were collected from *Orobanche* inflorescences parasitizing sunflower plants in the Jezreel Valley in northern Israel; *O. aegyptiaca* and *O. ramosa* seed were collected from *Orobanche* inflorescences parasitizing tomato and potato plants, respectively, on the Golan Heights in northern Israel; *O. cernua* seed were collected from *Orobanche* inflorescences parasitizing tomato in Beit Dagan in the central coastal plain of Israel. The inflorescences were dried in a greenhouse for 60 days and the seed were separated with 300- μ m sieves, then stored in the dark at 4°C. The germination potential of the seed was tested in the presence of GR24 (a synthetic strigol analog germination stimulant) at 10 ppm. After a preconditioning period of 12 days at 24°C, germination rates of seed of *O. cumana*, *O. aegyptiaca*, *O. ramosa*, and *O. cernua* were 88, 80, 60, and 75%, respectively.

Field studies. Experiments were started in April 1997 and April 1998 at the Volcani Center, Bet Dagan. The field was sprinkler irrigated to a depth of 60 cm, plowed, fertilized, and 1.93-m-wide beds were prepared. The soil was fumigated with methyl bromide at 500 kg/ha and was artificially infested, in two parallel furrow rows 90 cm apart, with *O. cumana*, *O. aegyptiaca*, *O. ramosa*, or *O. cernua* seed at a rate of 200 mg of seeds mixed with 16 ml of fine sand per meter row. The seed were placed at a furrow-row depth of 8 cm in 20-m-long plots. Half (10 m) of each plot was sown with R sunflower seed and the other half with S sunflower seed. Control plots without broomrape seed were sown with R and S sunflower cultivars as above. The sunflowers were sown manually and were

Corresponding author: H. Eizenberg
E-mail: eizenber@volcani.agri.gov.il

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thinned to 3 plants/m at the four- to six-true-leaf stage. *Orobanch* shoots were counted weekly throughout the season. Sunflower vigor was estimated visually using a scale of 0 (entirely dead plants) to 100 (healthy, green and vigorous plants) compared with the noninfested control plots. For example, estimation of 50% vigor indicated that a plant was stunted by approximately 50% compared with the noninfested control, or a plant that showed yellowing or withering symptoms, or a combination of those symptoms. The heads of the sunflower plants from each plot (20 m²) were collected and weighed after their seed had ripened.

Greenhouse studies. Experiments were conducted under greenhouse conditions at 21 and 29°C (night and day) with an average of 14 h of natural daylight. The experiments were conducted in 18-cm-diameter pots containing vermiculite and air-dried soil (medium-heavy clay-loam soil; 55% clay, 23% silt, 20% sand, 2% organic matter, pH 7.1; 1:1 vol/vol). Soil containing N:P:K (20:20:20, wt/wt/wt) commercial slow-release fertilizer at 1 g/kg soil, and *O. cumana*, *O. aegyptiaca*, *O. ramosa*, or *O. cernua* seed at 5 mg/kg soil were mixed in a cement mixer for 10 min. The control pots did not contain *Orobanch* seed.

Three to five seed of R or S sunflower were planted in each pot and thinned to one seedling per pot after emergence. Treatments included R or S sunflower plants grown in pots inoculated with each of the *Orobanch* spp., and a noninfested control. The pots were drip irrigated as needed. The *Orobanch* shoots that emerged above the soil were counted weekly. Sunflower vigor was estimated using a scale of 0 to 100, as above. Inhibition was characterized as withering and yellowing leaves, small plants, and small diameter of the sunflower inflorescence. At the end of the experiment, 90 days after planting, sunflower shoots were harvested, dried at 70°C for 48 h, and measured as a total dry sunflower biomass. The sunflower roots were washed of soil and healthy and degenerate parasites (3) attached to the roots were counted, dried at

70°C for 48 h, and measured as a total broomrape biomass.

Polyethylene bag studies. Polyethylene bags (PEB) were prepared according to Parker and Dixon (15) with modifications according to Eizenberg et al. (6). This method facilitates continuous observation of the host roots and monitoring of the various stages of *Orobanch* development and the parasitism process: seed germination, attachment of the germ tubes to the host roots, spider formation, and the appearance of parasite spikes.

R and S sunflower seedlings were germinated in a vermiculite medium in a growth chamber under 21 and 29°C (night and day) temperatures. At the cotyledon stage, 7 to 10 days after planting, the seedlings were transferred gently into PEB containing fiberglass filter paper (GF/A, Whatman) onto which sterilized *Orobanch* seed had been scattered. Two sunflower seedlings were placed in each bag, with each root system on half of each GF/A sheet, together with sterilized *O. cumana*, *O. aegyptiaca*, *O. ramosa*, or *O. cernua* seed. The plants were nourished with half-strength Hoagland nutrient solution (12).

Sunflower plants were grown in a growth chamber under 21 and 29°C (night and day) temperatures. The responses of R and S sunflower plants to the *Orobanch* spp. were expressed as an R/S ratio. This value was obtained for each stage of the *Orobanch* parasitism by dividing the percentage of germinated seed, attached germs, tubercle production, or degenerate parasites observed on the R sunflower roots by the number observed on the S roots. This treatment of the data enabled comparison of compatibility of the sensitive sunflower to each development stage of each of the *Orobanch* spp. with that of the resistant one. R and S sunflower plants were grown in PEB with no *Orobanch* seed as untreated controls. Observations were carried out with a binocular microscope (DZ, Zeiss, Germany) at ×40 magnification, in order to observe and monitor the events of seed germination and the successful continuation of the parasitism

process. Microscopic observations covered the area of the main sunflower root, on which the numbers of broomrape seed, germinating seed, tubercles, spikes, and degenerate attachments were recorded.

Experimental design and analysis.

The field study was arranged in a completely randomized split plot design, with five replications, with *Orobanch* spp. serving as a main plot and sunflower cultivars as a subplot. The greenhouse experiments were arranged in a completely randomized design (*Orobanch* spp. and sunflower cultivars) with six replications for each treatment. The cultivation in the PEB experiment was performed in a completely randomized design with two factors (*Orobanch* spp. and sunflower cultivar) and with eight replications for each treatment. The experiment was repeated twice. The results were subjected to analysis of variance by means of the JMP software (version 4.0.3; SAS Institute Inc., Cary, NC). Means were separated by least significant difference based on a Tukey Kremer Honestly Significant Different (HSD) test ($P \leq 0.05$).

RESULTS

Field study. The year–experiment interaction for the field experiments conducted in 1997 and 1998 was not significant; therefore, the results obtained from the two experiments were combined. Differences were observed in the response of R and S sunflower cultivars to *Orobanch* spp. infection (Table 1).

By 74 days after planting (DAP), the numbers of *Orobanch* shoots that emerged around R sunflower plants infected with *O. cumana*, *O. aegyptiaca*, and *O. ramosa* were 1.5 to 6 shoots per plant, but this infection level did not cause any damage to the R sunflower plants, which developed normally throughout the experiment. Moreover, broomrape shoots did not begin to emerge around the R plants until 60 DAP; whereas, around the S plants, *O. cumana* shoots began to emerge 32 DAP, and those of *O. aegyptiaca* and *O. ramosa* after 46 DAP. The infection levels in the S sunflower plants grown in the broomrape-

Table 1. *Orobanch* spp. parasitism and development of sunflowers of resistant cv. Ambar (R) and sensitive cv. Adi (S), grown in the field in spring 1997 and 1998

<i>Orobanch</i> spp.	Sunflower	<i>Orobanch</i> parasitism (Shoots/plant)				Sunflower development (%) ^b			
		32 DAP ^a	46 DAP	60 DAP	74 DAP	32 DAP	53 DAP	74 DAP	95 DAP
<i>O. cumana</i>	R	0	0	0.5	1.5	96	100	96	100
	S	4.6	8	24	45	100	100	72	42
<i>O. aegyptiaca</i>	R	0	0	1	6	100	96	98	98
	S	0	5	18	31	98	100	84	68
<i>O. ramosa</i>	R	0	0	2	5	100	98	96	98
	S	0	6	22	28	96	100	80	64
<i>O. cernua</i>	R	0	0	0	0	100	100	100	97
	S	0	0	0	0	100	97	96	100
LSD (0.05) ^c	...	1.8	3.2	6.7	9.4	5.8	5.3	7.1	9.3

^a DAP = days after planting. Data are means obtained from 10 replications in 2 years of experiments.

^b Sunflower development on a scale of 0 to 100, with 0 representing dead plants and 100 representing healthy plants, compared with the noninfested control.

^c Means were separated by least significant difference (LSD) based on Tukey-Kramer Honestly Significant Difference (HSD) test, ($P \leq 0.05$).

infested plots increased considerably in the course of time, and reached 28 to 45 *Orobanch* shoots per plant in the plots infested with *O. cumana*, *O. aegyptiaca*, and *O. ramosa*. *Orobanch* infection retarded the development of the S sunflower plants. The inhibition was characterized as withering and yellowing leaves. Inhibition was first observed 74 DAP and had increased by 95 DAP, when sunflower development in plots infested with *O. aegyptiaca*, *O. ramosa*, and *O. cumana* was 42, 68, and 64%, respectively, of that of the S plants in the noninfested control plots. The most severe damage was observed in the S sunflower plants grown in the *O. cumana*-infested plots, in which the damage was manifested as plant withering, stunting, and development of undersized sunflower inflorescences that produced empty kernels. *O. cernua* shoots did not emerge in either the R or the S sunflowers; therefore, no damage to the plants could be observed (Table 1).

Sunflower yield was measured in order to determine the effect of the parasitism on sunflower seed production. Seed production in *O. cumana*-infested S sunflowers was only 8% of that in the S plants grown

in the noninfested control plots (Fig. 1), and *O. aegyptiaca* and *O. ramosa* reduced seed production by 26 and 22%, respectively, in S plants compared with the controls. None of the three *Orobanch* spp. affected the seed production of the R sunflowers grown in the infested soil and their yields were the same as that of the plants grown in the noninfested plots. There also was no difference in seed production between the R and S sunflower plants grown in the *O. cernua*-infested and noninfested plots.

Greenhouse studies. There was no experiment-treatment interaction between the greenhouse experiments; therefore, the results of both experiments were combined. The R plants were not significantly infected by any of the tested *Orobanch* spp., in contrast with the high infection levels observed in the S plants in the pots inoculated with *O. cumana*, *O. aegyptiaca*, or *O. ramosa* (Table 2). On the S plants, *O. cumana* infection was first observed 32 DAP, and *O. aegyptiaca* and *O. ramosa* infections at 46 DAP. The infection levels of the S plants 74 DAP reached 26 to 48 inflorescences per plant, for *O. cumana*, *O. aegyptiaca*, and *O. ramosa*. No *O. cernua*

infection could be detected on the roots of either R or S plants throughout the experiments. The development of the infected S plants was significantly inhibited by each of the three *Orobanch* spp., whereas the same parasite species had no effect on the development of the R plants; the latter continued to develop normally in infested pots, with no difference from those growing in the noninfested pots (Table 2). *O. cumana* inhibited development of S plants more severely than did *O. aegyptiaca* or *O. ramosa*, as observed at 46 DAP. The vigor and appearance of the plants grown in the pots infested with *O. cumana* continued to deteriorate and, at 74 DAP, their development was evaluated at 24% of noninfested, compared with 50 and 45% for the plants grown in pots infested with *O. aegyptiaca* and *O. ramosa*, respectively. *O. cernua* did not affect either R or S sunflower plants, which continued to develop normally (Table 2).

Parasite attachments on the roots were counted at the end of the experiment. The proportions of degenerate attachments in the *O. cumana*-, *O. aegyptiaca*-, and *O. ramosa*-infected R sunflower roots were high, compared with a low proportion of degenerated attachments found on the S sunflower plant roots (Fig. 2): between 78 to 87% of the attachments found on the R sunflower plant roots grown in the pots infested with *O. cumana*, *O. aegyptiaca*, and *O. ramosa* were degenerate or dead, compared with 8 to 20% of those on the S plant roots in similar pots. No *O. cernua* attachments were observed on either the R or the S sunflower roots grown in the *O. cernua*-infested pots (Fig. 2).

The total dry biomass, including healthy and degenerate attachments as well as the shoots of each of the tested *Orobanch* spp. (*O. cumana*, *O. aegyptiaca*, and *O. ramosa*) on the roots of S sunflower plants was much higher than that of the same species collected from the roots of the R sunflower plants, and there was no difference among the *Orobanch* spp. (Fig. 3).

Differences were observed between the dry biomasses of R and S sunflower plants infected with *Orobanch* spp. (Fig. 4).

Fig. 1. Resistant (R) and susceptible (S) sunflower yields (percentage of noninfested control) grown under field conditions. Sunflowers were grown in spring 1997 and 1998. The data represent means of 10 replications from 2 years of experiments. Vertical bar represents least significant difference (LSD) based on Tukey-Kramer Honestly Significant Difference test ($P \leq 0.05$).

Table 2. *Orobanch* spp. parasitism and development of sunflowers of resistant cv. Ambar (R) and the sensitive cv. Adi (S), grown in pots in the greenhouse under controlled conditions of 21 and 29°C (night and day)

<i>Orobanch</i> spp.	Sunflower	<i>Orobanch</i> parasitism (Shoots/plant)				Sunflower development (%) ^b			
		32 DAP ^a	46 DAP	60 DAP	74 DAP	32 DAP	46 DAP	60 DAP	74 DAP
<i>O. cumana</i>	R	0	0	0.5	1.5	100	98	96	96
	S	4.1	11	26	48	100	64	40	24
<i>O. aegyptiaca</i>	R	0	0	0	5	96	96	100	96
	S	0	3.4	17.2	33	98	76	62	50
<i>O. ramosa</i>	R	0	0	0	3	96	98	100	100
	S	0	2.8	12	26	98	82	65	45
<i>O. cernua</i>	R	0	0	0	0	100	100	94	100
	S	0	0	0	0	97	100	97	99
LSD (0.05) ^c	...	1.2	2.4	5.9	7.5	5.6	8.3	7.7	8.6

^a DAP = days after planting. Data are means obtained from 12 replications in two experiments.

^b Sunflower development on a scale of 0 to 100, with 0 representing dead plants and 100 representing healthy plants, compared with the noninfested control.

^c Means were separated by least significant difference (LSD) based on Tukey-Kramer Honestly Significant Difference (HSD) test, ($P \leq 0.05$).

Whereas the R sunflower dry biomass was not influenced by any of the three *Orobanch* spp. infections, the dry weights of S plants infected with *O. cumana*, *O. aegyptiaca*, and *O. ramosa* were 24 to 54% of the dry weight of the noninfested controls (Fig. 4). No differences in dry weight were observed between the R and S sunflower plants grown in *O. cernua*-infested pots and those grown in noninfested control pots.

PEB studies. No experiment-treatment interaction between the experiments conducted in the PEB system was found; therefore, results of the experiments were combined. The developmental stages—germination, attachments, tubercle development, or tubercles mortality—of *O. cumana*, *O. aegyptiaca*, *O. ramosa*, and *O.*

cernua on the roots of the R and the S sunflower plants are summarized in Table 3. *Orobanch* seed germinated at rates of 67 to 92% for *O. cumana*, *O. aegyptiaca*, *O. ramosa*, and *O. cernua* on the R cultivar and at rates of 70 to 88% on the S cultivar. However, the R/S ratios in the germination stage were not significantly different from 1 for each of the four *Orobanch* spp.

No significant differences were observed among *O. cumana*, *O. aegyptiaca*, and *O. ramosa* in attachment rate as a percentage of the germinated seed (Table 3). The R/S ratios were not significantly different from unity at this stage, indicating that no differences exist between the R and S sunflower plants in the attachment rates of *O. cumana*, *O. aegyptiaca*, and *O. ramosa* to their roots. Few *O. cernua* at-

tachments were observed on either the R or S roots and R/S ratios were not computed for attachments and further parasitism stages. The *O. cernua* germ tubes that succeeded in reaching the roots formed an initial contact that soon ceased to develop and the *O. cernua* germs died.

The percent of healthy developed tubercles of *O. cumana*, *O. aegyptiaca*, and *O. ramosa* were greater on the S than on the R plant roots, as indicated by R/S ratios <1 (Table 3). However, the R/S ratio of mortalities of tubercles of the same *Orobanch* spp. was >1, indicating that more degenerate attachments were found on the R than on the S sunflower roots. The R/S ratio for tubercles mortality was greatest in *O. cumana* compared with *O. aegyptiaca* or *O. ramosa*, indicating more compatibility between the resistance mechanism of *O. cumana* and sunflower than *O. aegyptiaca* or *O. ramosa*.

DISCUSSION

The results of the present study indicate that the resistance response of *O. ramosa* on resistant sunflower is similar to the response observed in *O. cumana* and in *O. aegyptiaca* (3,6). However the response of *O. cernua* to R and S sunflower differs. The roots of both R and S cultivars induce seed germination of each of the four *Orobanch* spp., and no difference in *Orobanch* seed germination rate was observed between the R and the S sunflowers. These results suggest that the incompatibility between sunflower and *O. cernua* is not blocked at the *Orobanch* seed germination stage.

The attachments on the roots of the S sunflower cv. Adi continued to develop normally, producing inflorescences and viable seed of three of the *Orobanch* spp. tested. *O. cernua* germinated but failed to establish in either the S or R sunflower root tissues.

This study demonstrates that, in Israel, *O. cernua* differs from *O. cumana* in its host range. The incompatibility reaction between the host and *O. cernua* was evident in the early stage of parasitism, attempting penetration into the host root tissue. Our results support the findings of a previous study (13), which used RAPD marker techniques to prove that there were differences between these two *Orobanch* spp. In contrast to the mode of the sunflower-*O. cernua* incompatibility reaction, *O. ramosa* succeeded in penetrating both R and S sunflower roots, but the incompatibility reaction occurred only on the R roots. With *O. ramosa*, parasitism was limited at the tubercle stage or, in some cases, even later, at the stage of inflorescence apex formation.

O. ramosa successfully penetrated the host tissues, after which most of the attachments on the R sunflower roots degenerated and died. This resistant response was observed in the R sunflower cv. Am-

Fig. 2. Degenerate attachments (percentage of total attachments) of *Orobanch* spp. parasitizing resistant (R) and susceptible (S) sunflowers, 80 days after planting. Pots were kept under controlled conditions (21 and 29°C, night and day). Numbers in parentheses are total attachments. Vertical bar represents least significant difference (LSD) based on Tukey-Kramer Honestly Significant Difference test ($P \leq 0.05$).

Fig. 3. *Orobanch* spp. dry biomass in resistant (R) and susceptible (S) sunflowers in pots grown under greenhouse conditions. The data are the means of 12 replications from two experiments. Vertical bar represents least significant difference (LSD) based on Tukey-Kramer Honestly Significant Difference test ($P \leq 0.05$).

bar in the three *Orobanch* spp. that parasitized sunflower, but not with *O. cernua*.

Previous studies on the resistance of sunflower to *Orobanch* spp. addressed the resistance mechanisms to *O. cumana* races A-C (1). The mechanisms are described as hypersensitivity and cortex necrosis (Or_2), involvement of *Orobanch*-toxic phenolic compounds, and condensation of cell walls by lignification (Or_3) (5,8,10,14). Involvement of 7-hydroxylated simple coumarins in the resistant response of sunflower to *O. cernua* has been proposed (17). Histological studies on the roots of *Orobanch* spp.-resistant purple vetch showed that the haustorium penetrated directly through the epidermis and cortex but stopped at the endodermis cells (8,10). Most of those studies discuss resistance mechanisms that are initiated shortly after the parasite penetrated the host root tissue. The resistant reaction described in the present study is different. Parasite mortality occurs at a later stage of the parasitism process (e.g., at the tubercles stage or even at shoot formation and emergence). The resistant response of cv. Ambar toward *O. ramosa* was found in this study to be similar to the response to *O. cumana* and *O. aegyptiaca*; namely, degeneration and death of the parasite after successful germination, attachment, penetration, and establishment.

It can be concluded that this type of response is a general characteristic of the resistance mechanism of Ambar regardless of the *Orobanch* spp., except in the case of *O. cernua*.

Others researchers have proposed that the resistance mechanism of sunflower type Or_4 or Or_5 is a result of increased phytoalexins or phytoalexins production (1,17). It can be hypothesized that the resistance mechanism of the sunflower cv. Ambar also may be explained by resistance types Or_4 or Or_5 (5).

Field studies showed that *O. cumana* virulence on the sensitive cultivar was higher than the virulence of *O. aegyptiaca* and *O. ramosa*, resulting in severe damage and yield reduction in *O. cumana*-infested fields. The resistant sunflower Ambar and the susceptible sunflower Adi were selected for this study due to their resistance and susceptibility, respectively, only to *O. cumana*. Thus, it might explain the high infection level due to the high compatibility between the sensitive sunflower and *O. cumana*.

The present study has identified the pathogenicity and virulence potential of *O. ramosa* and *O. cernua* to Ambar and Adi sunflower cultivars. The results have enabled us to include resistant sunflower cultivars in crop rotations in *O. cumana*-,

O. aegyptiaca-, and *O. ramosa*-infested fields in Israel. The findings of this study also enabled us to recommend that sunflower growers plant resistant and sensitive sunflower cultivars in *O. cernua*-infested fields. This broomrape species may serve as a "trap crop" (11) in order to reduce broomrape soil seed bank.

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Fig. 4. Resistant 'Ambar' and sensitive 'Adi' sunflower dry weights (percentage of those of noninfested sunflower plants) grown in pots infested with *Orobanch* spp. under controlled greenhouse conditions. The data are the means of 12 replications from two experiments. Vertical bar represents least significant difference (LSD) based on Tukey-Kramer Honestly Significant Difference test ($P \leq 0.05$).

Table 3. *Orobanch* spp. development at difference parasitism stages on resistant (R) and susceptible (S) sunflower cultivars

<i>Orobanch</i> spp.	Germination (% of seed)			Attachments (% of germs)			Tubercles (% of attachments)			Mortality (% of tubercles)		
	R	S	R/S	R	S	R/S	R	S	R/S	R	S	R/S
<i>O. cumana</i>	85 ^a	87	0.98	36	37	0.97	27	82	0.33	93	14	6.64
<i>O. aegyptiaca</i>	74	72	1.03	38	40	0.95	25	81	0.30	88	20	4.4
<i>O. ramosa</i>	67	70	0.96	30	32.4	0.93	21	72	0.29	85	22	3.86
<i>O. cernua</i>	92	88	1.05	1.2	0.8	N/A	N/A	N/A	N/A	N/A	N/A	N/A
LSD (0.05) ^b	8.7	7.5	0.14	9.3	9.1	0.08	8.2	12.3	0.09	9.0	8.7	0.74

^a Data were arcsine transformed but the true values are presented. R/S = ratio of the rate of each developmental stage for the susceptible cultivar divided by the resistant cultivar; N/A = no *O. cernua* tubercles were observed.

^b Means were separated by least significant difference (LSD) based on Tukey-Kramer Honestly Significant Difference (HSD) test, ($P \leq 0.05$).

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